Agilent Solutions for Analyzing Polycyclic Aromatic Hydrocarbons in Seafood

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PAH analytical method summary

- **NOAA** NMFS-NWFSC-59, 2004 **GPC + GC/MS** is the current method specified for PAH analysis in seafood. It is a complex and time-consuming method so many customers are looking for an alternative.
- **QuEChERS + GC/MS** or **GC/MS/MS PAH analyzers** are the proposed solutions from Agilent to simplify sample preparation, reduce cycle time, and provide increased sensitivity. Backflush, Multi Mode Inlet, and MS/MS are the technologies contributing to the performance and sample through-put.
- **QuEChERS + HPLC/Fluorescence** is an excellent screening tool. This Agilent solution was chosen by the US Food Emergency Response Network.

Why QuEChERS for Sample Prep?

When Compared to traditional sample prep methods:

- 25-50%+ time savings
- Reduced solvent usage: 10-15 mL/sample
- No chlorinated solvents required
- Extract multiple families of compounds with one extraction method
- Does not require advanced sample preparation experience

QuEChERS Seafood Extraction Method:

Step1:Extraction

Finfish, shellfish

↓

Weigh 3 g into 50 mL tube,
add 2 ceramic homogenizers

↓

Add 12 mL of water, vortex 30 sec

↓

Add 15 mL of ACN (1% AA), vortex 1 min

↓

Vertically shake for
1 min, centrifuge at 4000 rpm for 5 min

↓

Aspirate and transfer 8 mL of extract
To dispersive SPE (fatty sample)

Step2: d-SPE (dispersive-SPE)

Transfer 8 mL of extract from Step 1
To d-SPE (fatty sample)

↓

Vortex 1 min

↓

Centrifuge 400 rpm, 5 min

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Aspirate extract , filter through 0.45 um
Nylon filter, transfer to GC or HPLC Vial

↓

Step 3: Analyze
GC/MS or GC/QQQ
LC/UV/FLD or LC/QQQ
(require dilution with water 1:4 or 1:5
prior to LC)

QuEChERS: PAH Determination in Fish

Amenable to both GC and LC, UV/FLD and MS detection

Note: Solvent exchange is not required for GC analysis

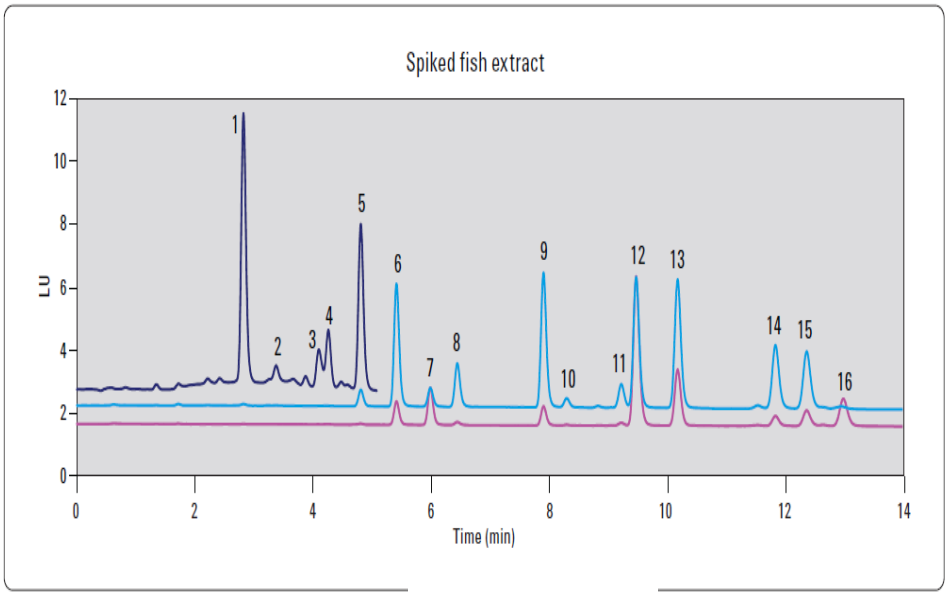
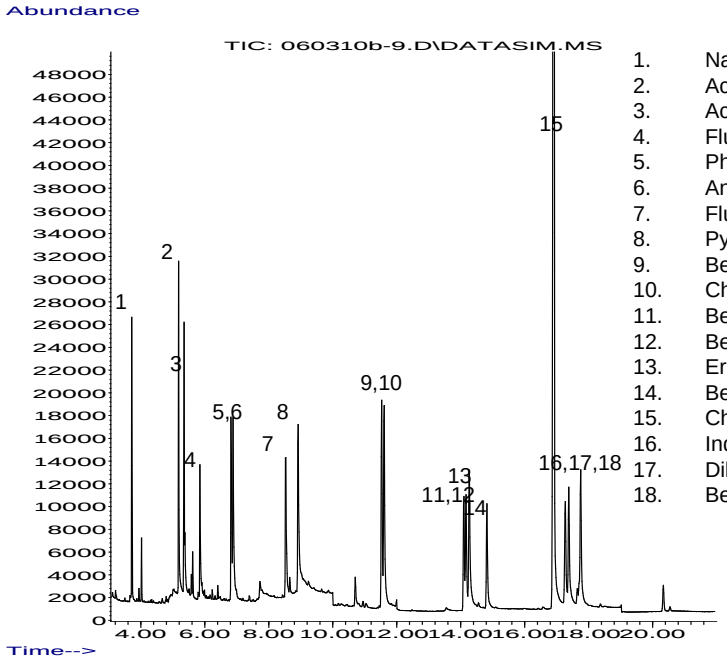


Figure 5. Overlay HPLC – FLD chromatograms of the spiked fish sample containing: 1. Nap 2. Acy 3. Ace 4. Flu 5. Phe 6. Ant 7. Fln 8. Pyr 9. BaA 10. Chr 11. BeP 12. BeA 13. BkF 14. DahA 15. BghiP 16. InP. The spiking level for this sample was level 1 (see Table 3). The blue portion of the chromatogram used the following excitation/emission wavelengths: 260-nm/352-nm; the red portion 260-nm/420-nm; the light blue portion: 260-nm/440-nm. For acenaphthylene, UV detection at 230-nm was used. Chromatographic conditions are shown in Table 1.



1. Napthalene
2. Acenaphthylene
3. Acenaphthene
4. Fluorene
5. Phenanthrene
6. Anthracene
7. Fluoranthene
8. Pyrene
9. Benz[a]anthracene
10. Chrysene
11. Benzo[b]fluoranthene
12. Benzo[k]fluoranthene
13. Erucylamide
14. Benz[a]pyrene
15. Cholesterol
16. Indeno[1,2,3-c,d]pyrene
17. Dibenz[a,h]anthracene
18. Benzo[g,h,i]perylene

50ppb EPA PAHs extracted from Swai fish using QuEChERS
DB-5ms 20m 0.18mm 0.18µmGC/MS SIM TIC

PAH Analysis: GC/MS with Column Backflush

-- Improved reliability and speed

Oven Program
50 °C for 0.8 min
then 70 °C/min to 180 °C for 0 min; then 7 °C/min to 230 °C for 1 min
then 40 °C/min to 280 °C for 1 min; then 25 °C/min to 335 °C for 3 min

Run Time 18.25 min

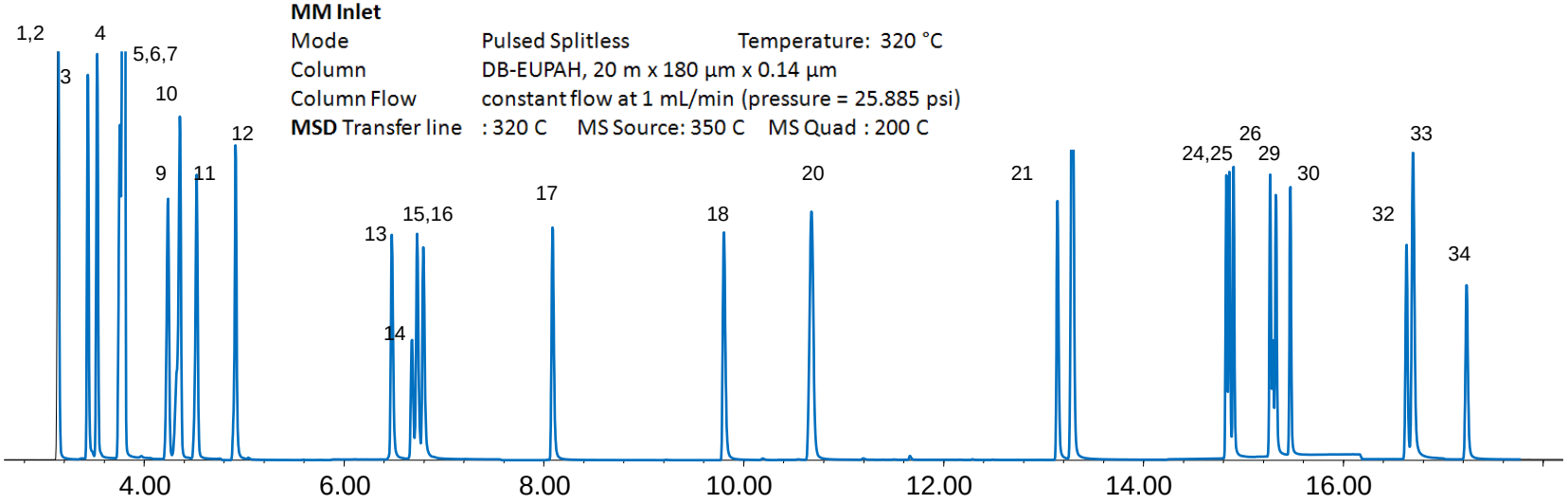
MM Inlet

Mode Pulsed Splitless Temperature: 320 °C

Column DB-EUPAH, 20 m x 180 µm x 0.14 µm

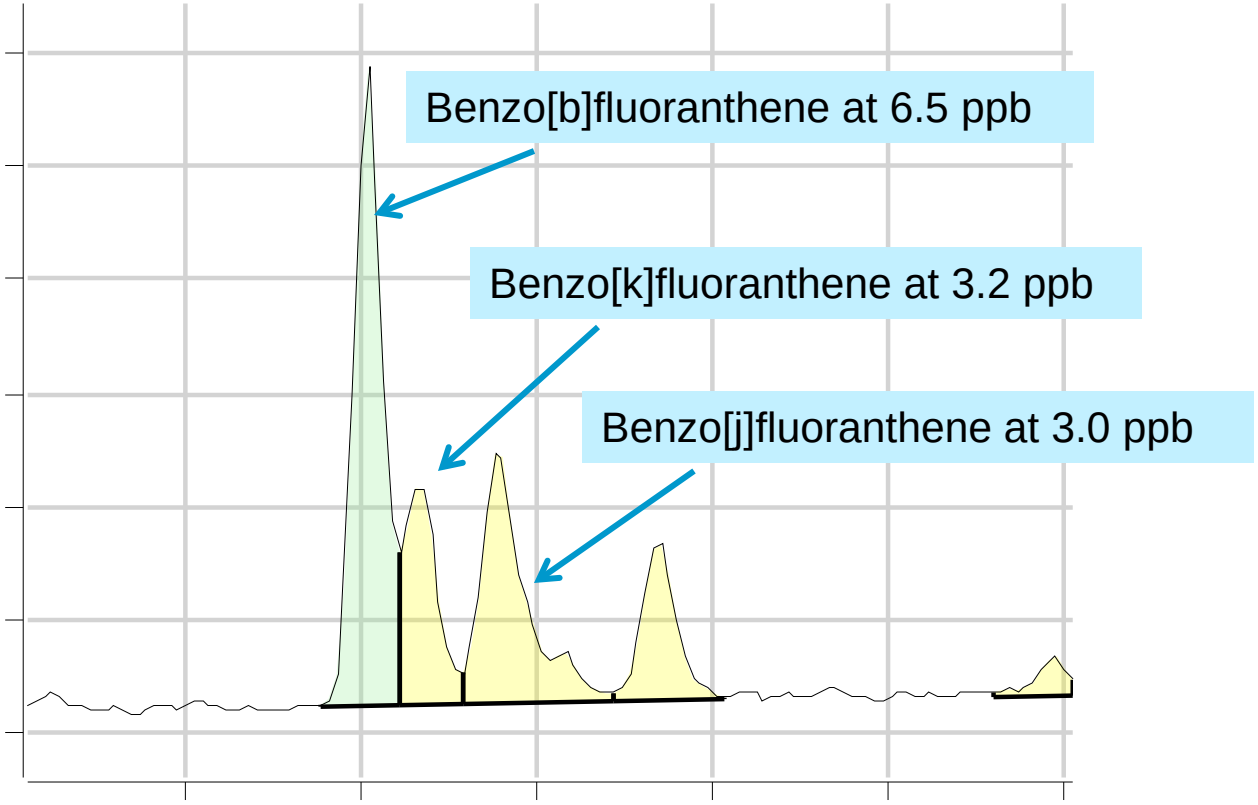
Column Flow constant flow at 1 mL/min (pressure = 25.885 psi)

MSD Transfer line : 320 C MS Source: 350 C MS Quad : 200 C



	Internal Std						
1	Naphthalene-d8	4	2-Methylnaphthalene	15	Phenanthrene	25	Benzo[k]fluoranthene
9	Acenaphthene-d10	5	Biphenyl	16	Anthracene	29	Benzo[a]pyrene
14	Phenanthrene-d10	6	2,6-dimethylnaphthalene	17	1-methylphenanthrene	30	Perylene
26	Benzo[a]pyrene-d12	7	HMB	18	Fluoranthene	32	Dibenz[a,h]anthracene
		8	Acenaphthylene	20	Pyrene	33	Indeno[1,2,3-cd]pyrene
		10	Acenaphthene	21	Benz[a]anthracene	34	Benzo[ghi]perylene
	Target Compounds	11	2,3,5-trimethylnaphtha...	22	Triphenylene		
2	Naphthalene	12	Fluorene	23	Chrysene		
3	1-methylnaphthalene	13	Dibenzothiophene	24	Benzo[b]fluoranthene		

Low detection limit with GC/MS/MS

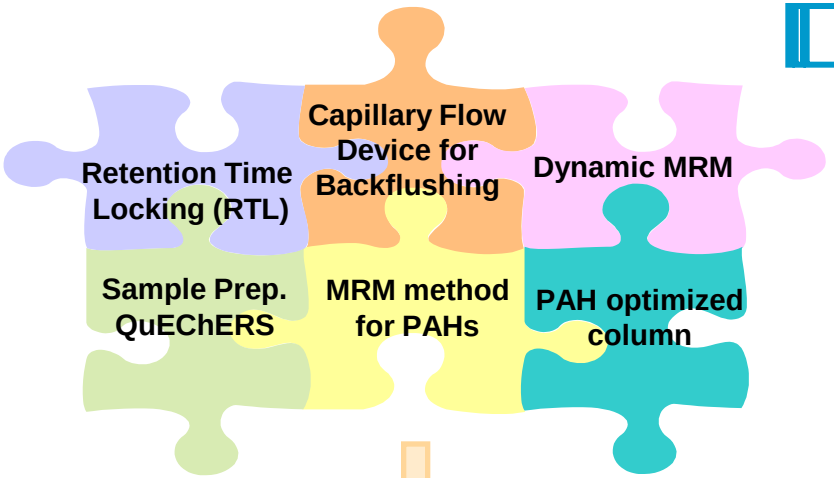


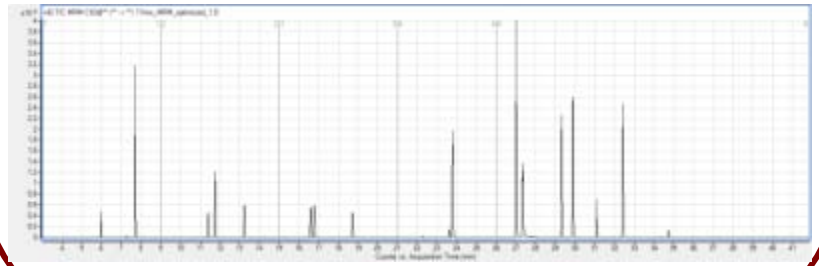
NIST Certified Reference Material 1974b Mussel sample

Ready to use PAH Analyzers

configured and tested at the factory for fast lab start-up

At Factory





Plug and Play

At User's Lab

Ready for the 1st run

Solutions from Agilent for PAH Analysis in Seafood

-- for higher productivity, better performance on Day ONE

- **QuEChERS:** ***simple and fast*** extraction and sample clean-up
- **GC/MS with Backflush:** using innovative Capillary Flow Technology ***shortens GC cycle time*** while keeping column and source cleaner longer!
- **GC/MS/MS with Backflush:** highest ***sensitivity*** for PAH analysis .
- **PAH Optimized Column:** ***optimized PAH separation*** in shortest cycle time
- **Factory setup and tested analyzers:** Reduces method development time!
- **HPLC Fluorescence** - sensitive quick screening method